

**This listing of claims will replace all prior versions and listings of claims in the application.**

**LISTING OF CLAIMS**

1-25. (Canceled)

26. (Currently Amended) A method for investigating a body fluid from a human subject having or suspected of having cancer for disseminated cancer cells, which comprises:

- obtaining a cell-containing fraction from the body fluid with enrichment of cancer cells and determining in the cell-containing fraction the expression of at least 2 genes which are selected from the group consisting of

- i) a human manganese superoxide dismutase **genes gene**;

- ii) a human thioredoxin reductase 1 **genes gene**; and

- iii) a human glutathione peroxidase 1 **genes gene**;

- providing a further cell-containing fraction of the body fluid from the same individual and determining the expression of the genes in the further cell-containing fraction; and

- comparing the expression for each of said at least 2 genes in the cell-containing fraction with its expression in the further cell-containing fraction, and

wherein the body fluid is selected from blood and bone marrow and an elevated expression of at least one of said at least 2 genes determined in the cell-containing fraction, as compared to its expression in the further cell-containing fraction, indicates the presence of disseminated cancer cells in the body fluid.

27. (Previously Presented) The method as claimed in claim 26, wherein the expression of a manganese superoxide dismutase gene, of a thioredoxin reductase 1 gene and of a glutathione peroxidase 1 gene is determined.

28. (Previously Presented) The method as claimed in claim 26, wherein the body fluid is blood.

29. (Previously Presented) The method as claimed in claim 26, wherein obtaining the cell-containing fraction from the body fluid with enrichment of cancer cells comprises

passing the body fluid or a cell-containing fraction thereof through a screen with a mesh or pore width of about 10 to 200  $\mu\text{m}$  and obtaining the cell fraction retained on the screen.

30. (Previously Presented) The method as claimed in claim 26, wherein the expression of a manganese superoxide dismutase gene and of at least one further gene selected from a thioredoxin reductase 1 genes and a glutathione peroxidase 1 genes is determined.

31. (Cancelled)

32. (Previously Presented) The method as claimed in claim 26, wherein the elevated expression of at least one of said genes indicates the presence of a tumor.

33. (Cancelled)

34. (Previously Presented) The method as claimed in claim 26, wherein the elevated expression of at least one of said genes indicates a risk to develop a metastasis or a recurrence.

35. (Cancelled)

36. (Previously Presented) The method as claimed in claim 26, wherein the manganese superoxide dismutase gene encodes a protein having the amino acid sequence of SEQ ID NO:13 or an allelic variant thereof.

37. (Previously Presented) The method as claimed in claim 26, wherein the manganese superoxide dismutase gene encodes an mRNA which is capable of being amplified using the primer sequences as set forth in SEQ ID NO:1 and SEQ ID NO:2.

38. (Previously Presented) The method as claimed in claim 26, wherein the thioredoxin reductase 1 gene encodes a protein having the amino acid sequence of SEQ ID NO:15 or an allelic variant thereof.

39. (Previously Presented) The method as claimed in claim 26, wherein the thioredoxin reductase 1 gene encodes an mRNA which is capable of being amplified using the primer sequences as set forth in SEQ ID NO:4 and SEQ ID NO:5.

40. (Previously Presented) The method as claimed in claim 26, wherein the human glutathione peroxidase 1 gene encodes a protein having the amino acid sequence of SEQ ID NO:17 or an allelic variant thereof.

41. (Previously Presented) The method as claimed in claim 26, wherein the human glutathione peroxidase 1 gene encodes an mRNA which is capable of being amplified using the primer sequences as set forth in SEQ ID NO:7 and SEQ ID NO:8.

42. (Previously Presented) The method as claimed in claim 26, wherein determining the expression of the at least 2 genes comprises determining mRNA expressed by the gene.

43. (Previously Presented) The method as claimed in claim 26, wherein the enrichment of cancer cells comprises isolating cancer cells.

44. (Previously Presented) The method as claimed in claim 26, wherein cancer cells are isolated by immunospecific adsorption, microdissection, by means of a density gradient or by filtration.

45. (Currently Amended) The method as claimed in claim 26, wherein the body fluid is blood and the cell-containing fraction is derived from blood and comprises the further cell-containing fraction comprise mononuclear cells.

46. (Previously Presented) The method as claimed in claim 29, wherein the screen has a mesh or pore width of about 20  $\mu$ m.

47. (Currently Amended) The method as claimed in claim 26, wherein the elevated expression of at least one of said at least 2 genes in the cell-containing fraction as compared to its expression in the further cell-containing fraction indicates the presence of disseminated cancer cells in the body fluid if the ratio of its expression ~~from~~ in the cell-containing fraction to its expression in the further cell-containing fraction is significantly higher than the corresponding average ratio of its expression in subjects not having cancer.

48. (Currently Amended) A method for investigating a body fluid from a human subject having or being suspected of having cancer for disseminated cancer cells, which comprises:

- obtaining a cell-containing fraction from the body fluid by passing the body fluid or a cell-containing fraction thereof through a screen with a mesh or pore width of about 20  $\mu$ m and obtaining the cell fraction retained on the screen;

- determining in the cell-containing fraction the expression of

- i) a human manganese superoxide dismutase gene which encodes an mRNA which is capable of being amplified using the primer sequences as set forth in SEQ ID NO:1 and SEQ ID NO:2, wherein the manganese superoxide dismutase gene encodes a protein having an amino acid sequence as set forth in SEQ ID NO:13 or an allelic variant thereof;
- ii) a human thioredoxin reductase 1 gene which encodes an mRNA which is capable of being amplified using the primer sequences as set forth in SEQ ID NO:4 and SEQ ID NO:5, wherein the thioredoxin reductase 1 gene encodes a protein having an amino acid sequence as set forth in SEQ ID NO:15 or an allelic variant thereof; and
- iii) a human glutathione peroxidase 1 gene which encodes an mRNA which is capable of being amplified using the primer sequences as set forth in SEQ ID NO:7 and SEQ ID NO:8, wherein the human glutathione peroxidase 1 gene encodes a protein having an amino acid sequence as set forth in SEQ ID NO:17 or an allelic variant thereof;

- providing a further cell-containing fraction of the body fluid from the same individual and determining the expression of the genes in the further cell-containing fraction; and

- comparing the expression for each of said at least 2 genes in the cell-containing fraction with its expression in the further cell-containing fraction, and

wherein the body fluid is selected from blood and bone marrow and an elevated expression of at least one of said at least 2 genes in the cell-containing fraction as compared to its expression in the further cell-containing fraction indicates the presence of disseminated cancer cells in the body fluid if the ratio of its expression ~~from~~ in the cell-containing fraction to its expression in the further cell-containing fraction is significantly higher than the corresponding average ratio of its expression in subjects not having cancer.

49. (Previously Presented) The method as claimed in claim 48, wherein the body fluid is blood and the cell-containing fraction and the further cell-containing fraction comprise mononuclear cells and wherein determining the expression of the at least 2 genes comprises determining mRNA expressed by the gene.

50. (Currently Amended) A method for investigating a body fluid from a human subject having or being suspected of having cancer for disseminated cancer cells, which comprises:

- obtaining a cell-containing fraction from the body fluid and determining in the cell-containing fraction the expression of at least 2 genes which are selected from the group consisting of

- i) a human manganese superoxide dismutase gene;

- ii) a human thioredoxin reductase 1 gene; and

- iii) a human glutathione peroxidase 1 gene; and

- comparing the expression for each of said at least 2 genes in the cell-containing fraction with its average expression in subjects not having cancer, and

wherein the body fluid is selected from blood and bone marrow and a significantly higher expression of at least one of said at least 2 genes in the cell-containing fraction as compared to its average expression in subjects not having cancer indicates the presence of

disseminated cancer cells in the body fluid.

51. (Previously Presented) The method as claimed in claim 50, wherein the expression of a manganese superoxide dismutase gene and of at least one further gene selected from a thioredoxin reductase 1 gene and a glutathione peroxidase 1 gene.

52. (Previously Presented) The method as claimed in claim 50, wherein the expression of a manganese superoxide dismutase gene, of a thioredoxin reductase 1 gene and of a glutathione peroxidase 1 gene is determined.

53. (Previously Presented) The method as claimed in claim 50, wherein the cell-containing fraction is derived from blood and comprises mononuclear cells.

54. (Previously Presented) The method as claimed in claim 50, wherein determining the expression of the at least 2 genes comprises determining mRNA expressed by the gene.

55. (Previously Presented) The method as claimed in claim 50, wherein the manganese superoxide dismutase gene encodes a protein having an amino acid sequence as set forth in SEQ ID NO:13 or an allelic variant thereof.

56. (Previously Presented) The method as claimed in claim 50, wherein the manganese superoxide dismutase gene encodes an mRNA which is capable of being amplified using the primer sequences as set forth in SEQ ID NO:1 and SEQ ID NO:2.

57. (Previously Presented) The method as claimed in claim 50, wherein the thioredoxin reductase 1 gene encodes a protein having an amino acid sequence as set forth in SEQ ID NO:15 or an allelic variant thereof.

58. (Previously Presented) The method as claimed in claim 50, wherein the thioredoxin reductase 1 gene encodes an mRNA which is capable of being amplified using the primer sequences as set forth in SEQ ID NO:4 and SEQ ID NO:5.

59. (Previously Presented) The method as claimed in claim 50, wherein the human glutathione peroxidase 1 gene encodes a protein having an amino acid sequence as set forth in SEQ ID NO:17 or an allelic variant thereof.

60. (Previously Presented) The method as claimed in claim 50, wherein the human glutathione peroxidase 1 gene encodes an mRNA which is capable of being amplified using the primer sequences as set forth in SEQ ID NO:7 and SEQ ID NO:8.

61. (Cancelled)

62. (Previously Presented) The method as claimed in claim 50, wherein the elevated expression of at least one of said genes indicates the presence of a tumor.

63. (Previously Presented) The method as claimed in claim 44, which is for diagnosis of a tumor.

64. (Previously Presented) The method as claimed in claim 50, wherein the elevated expression of at least one of said genes indicates a risk to develop a metastasis or a recurrence.

65. (Previously Presented) The method as claimed in claim 46, which is for estimating the risk to develop a metastasis or a recurrence.

66. (Currently Amended) A method for investigating a body fluid for disseminated cancer cells in a human subject having or being suspected of having cancer, which comprises:

- obtaining a cell-containing fraction from the body fluid and determining in the cell-containing fraction the expression of

- i) a human manganese superoxide dismutase gene which encodes an mRNA which is capable of being amplified using the primer sequences as set forth in SEQ ID NO:1 and SEQ ID NO:2, wherein the manganese superoxide dismutase gene encodes a protein having an

amino acid sequence as set forth in SEQ ID NO:13 or an allelic variant thereof;

- ii) a human thioredoxin reductase 1 gene which encodes an mRNA which is capable of being amplified using the primer sequences as set forth in SEQ ID NO:4 and SEQ ID NO:5, wherein the thioredoxin reductase 1 gene encodes a protein having an amino acid sequence as set forth in SEQ ID NO:15 or an allelic variant thereof; and
- iii) a human glutathione peroxidase 1 gene which encodes an mRNA which is capable of being amplified using the primer sequences as set forth in SEQ ID NO:7 and SEQ ID NO:8, wherein the human glutathione peroxidase 1 gene encodes a protein having an amino acid sequence as set forth in SEQ ID NO:17 or an allelic variant thereof;  
and

- comparing the expression for each of said at least 2 genes in the cell-containing fraction with its average expression in subjects not having cancer,

wherein the body fluid is selected from blood and bone marrow and a significantly higher expression of at least one of said at least 2 genes in the cell-containing fraction as compared to its average expression in subjects not having cancer ~~to indicate~~ indicates the presence of disseminated cancer cells in the body fluid.

67. (Previously Presented) The method as claimed in claim 66, wherein the body fluid is blood and the cell-containing fraction comprises mononuclear cells and wherein determining the expression of the at least 2 genes comprises determining mRNA expressed by the gene.

68. (Currently Amended) A method for investigating a blood or bone marrow sample for disseminated cancer cells in a human subject having or suspected of having cancer, comprising:

(a) obtaining a blood or bone marrow sample from the human subject having or suspected of having cancer to obtain a test fraction;



(b) obtaining a blood or bone marrow sample from a healthy human subject not suffering from cancer to obtain a reference fraction;

(c) isolating mRNA from the test fraction and reference fraction to obtain an mRNA test sample and an mRNA reference sample, respectively;

(d) measuring the expression level of human manganese superoxide dismutase (MNSOD), human thioredoxin reductase 1 (TXNRD1), and human glutathione peroxidase I (GPX1) in the mRNA sample and the mRNA reference sample, wherein the measuring is by reverse transcription and PCR with primers selected from the nucleotides of SEQ ID NOs: 1 and 2 for MNSOD; SEQ ID NOs: 3 and 4 for ~~TXNRD1~~ TXNRD1; and SEQ ID NOs: 7 and 8 for GPX1; and

(e) comparing the expression of MNSOD, ~~TXNRD1~~ TXNRD1, and GPX1 in the mRNA test sample to the mRNA reference sample, and wherein a significantly higher expression of MNSOD, ~~TXNRD1~~ TXNRD1, and GPX1 in the mRNA test sample as compared to the mRNA test reference sample indicates the presence of disseminated cancer cells in the blood or bone marrow sample from the human subject having or suspected of having cancer.

69. (New) A method for investigating a body fluid from a human subject having or being suspected of having cancer for disseminated cancer cells, which comprises:

- obtaining a cell-containing fraction from the body fluid with enrichment of cancer cells;

- determining in the cell-containing fraction the expression of

i) a human manganese superoxide dismutase gene which encodes an mRNA which is capable of being amplified using the primer sequences as set forth in SEQ ID NO:1 and SEQ ID NO:2, wherein the manganese superoxide dismutase gene encodes a protein having an amino acid sequence as set forth in SEQ ID NO:13 or an allelic variant thereof;

ii) a human thioredoxin reductase 1 gene which encodes an mRNA which is capable of being amplified using the primer sequences as set forth in SEQ ID NO:4 and SEQ ID NO:5, wherein the thioredoxin reductase 1 gene encodes a protein having an amino acid sequence as set forth in SEQ ID NO:15 or an allelic variant thereof; and

iii) a human glutathione peroxidase 1 gene which encodes an mRNA which is capable of being amplified using the primer sequences as set forth in SEQ ID NO:7 and SEQ ID NO:8, wherein the human glutathione peroxidase 1 gene encodes a protein having an amino acid sequence as set forth in SEQ ID NO:17 or an allelic variant thereof;

- providing a further cell-containing fraction of the body fluid from the same individual and determining the expression of the genes in the further cell-containing fraction; and

- comparing the expression for each of said at least 2 genes in the cell-containing fraction with its expression in the further cell-containing fraction, wherein the body fluid is blood and the cell-containing fraction and the further cell-containing fraction comprise mononuclear cells;

wherein determining the expression of the at least 2 genes comprises determining mRNA expressed by the gene; and

wherein an elevated expression of at least one of said at least 2 genes in the cell-containing fraction as compared to its expression in the further cell-containing fraction indicates the presence of disseminated cancer cells in the body fluid if the ratio of its expression from the cell-containing fraction to the further cell-containing fraction is significantly higher than the average ratio of its expression in subjects not having cancer.

70. (New) A method for investigating a body fluid from a human subject having or being suspected of having cancer for disseminated cancer cells, which comprises:

- obtaining a cell-containing fraction from the body fluid with enrichment of cancer cells;

- determining in the cell-containing fraction the expression of

i) a human manganese superoxide dismutase gene which encodes an mRNA which is capable of being amplified using the primer sequences as set forth in SEQ ID NO:1 and SEQ ID NO:2, wherein the manganese superoxide dismutase gene encodes a protein having an amino acid sequence as set forth in SEQ ID NO:13 or an allelic variant thereof;

ii) a human thioredoxin reductase 1 gene which encodes an mRNA which is capable of being amplified using the primer sequences as set forth in SEQ ID NO:4 and SEQ

ID NO:5, wherein the thioredoxin reductase 1 gene encodes a protein having an amino acid sequence as set forth in SEQ ID NO:15 or an allelic variant thereof; and

iii) a human glutathione peroxidase 1 gene which encodes an mRNA which is capable of being amplified using the primer sequences as set forth in SEQ ID NO:7 and SEQ ID NO:8, wherein the human glutathione peroxidase 1 gene encodes a protein having an amino acid sequence as set forth in SEQ ID NO:17 or an allelic variant thereof; and

- comparing the expression for each of said at least 2 genes in the cell-containing fraction with its average expression in subjects not having cancer, wherein the body fluid is blood and the cell-containing fraction comprises mononuclear cells;

wherein determining the expression of the at least 2 genes comprises determining mRNA expressed by the gene; and

wherein a significantly higher expression of at least one of said at least 2 genes in the cell-containing fraction as compared to its average expression in subjects not having cancer indicates the presence of disseminated cancer cells in the body fluid.